

iGEM TU/e 2014

Biomedical Engineering

Eindhoven University of Technology
Room: Ceres 0.04
Den Dolech 2, 5612 AZ Eindhoven
The Netherlands
Tel. no. +31 50 247 55 59
2014.igem.org/Team:TU_Eindhoven

Date

11 August 2014

FACS - DBCO-PEG 10 kDa

Table of contents

Title	1	Stock solutions	3
FACS - DBCO-PEG 10 kDa	2	Preparation of FACS samples	3

1 Stock solutions

- 300 μ M DBCO-PEG 10 kDa in DMSO
- Buffer: PBS

2 Preparation of FACS samples

- Prepare following tubes:

Tube	[DBCO]	Cells (10^9)	DBCO volume to add (μ L) (300 μ M)	DBCO/tag ratio
1	0	200 μ L		
2	30 μ M	200 μ L	22.2	300.5

- Make sure you vortex the cell well before and after adding the DBCO-PEG
- React DBCO tubes for 1h to 6h in shaking block at 4°C and 300rpm
- Prepare FACS samples after 1h and 6h:
 - Spin down the cells for 10 min at 13,400 rpm and discard the supernatant
 - Resuspend with 1 mL ice cold PBS
 - Spin down the cells for 10 min at 13,400 rpm and discard the supernatant
 - Put on ice until FACS
 - Right before FACS: resuspend with 200 μ L ice cold PBS